AN OLFACTOMETER PERMITTING STIMULUS SPECIFICATION IN MOLAR TERMS*

F. Nowell Jones
University of California, Los Angeles

Thresholds for chemoreceptors - olfactory or gustatory - are best given in terms of the molecular concentration of stimulating substance in the medium used to convey it to the receptor surface. This implies that control must be exercised over all other stimulus variables. In the case of olfaction, these are stimulus blast duration, pressure, volume, temperature, and humidity. Some of the previous attempts to accomplish this control have been reviewed by Wenzel,[1] and the difficulty encountered when an aspect of the stimulus other than concentration is used as a threshold definition has been demonstrated elsewhere.[2]

Difficulties of design center around two main problems, first, accuracy in specifying stimulus concentration, and, second, cost. Having a quantity of pure air with a known stimulus concentration, it is relatively easy to devise a means of delivering a controlled blast of it to S's nose, although doing it inexpensively is not too easy. The solution to these simultaneous problems to be presented below is not the only one possible, but it has proved simple and relatively uncumbersome.

Insert Fig. 1 about here

Figure 1 diagrams the apparatus. The odorous stimuli are contained in a series of evaporation bottles. Each unit, A, really consists of two bottles containing identical materials, so that as odorous air is drawn off, dilution progresses slowly, and each bottle has a glass wool "wick" to hasten

evaporation. In each absorption unit, the odorous material is dissolved in mineral oil, so that the odorous air is not too far above the expected threshold concentration. When necessary to cover the range of individual differences, two or three different dilutions may be prepared. Since there is no bubbling of air, there is no chance of aerosol formation. For each odor a syringe, B, three-way stopcock, C, and nosepiece, D, are provided. These are clamped into holders on the main body of the apparatus, and connected to the absorption bottles by means of a short length of silicone rubber tubing.[3] All connections among parts are made with silicone rubber, and all rubber stoppers are covered with aluminum foil. The nosepieces are especially fabricated, and provide air-tight connections to the nostril. When clamped in position, the plunger of the syringe makes contact with a rack, E, which is driven by a constant-speed motor, F, controlled by a switch, G. A stop, H, and the length of the toothed portion of the rack determine respectively the beginning point of the movement of the rack, and its termination. Since the stop and the motor mount are adjustable, the extent of movement, and therefore the amount of air forced from the syringe may be readily varied. As presently geared, a blast of 40 cc. is delivered in 0.8 sec. These magnitudes are such as to minimize quantity of stimulus blast as a determiner of threshold. The absorption units are stored in a constant temperature bath and are immersed in a constant temperature bath during experimentation. The temperature of the baths is 25.9° C. with a maximum variation of ± 0.1° C. Pure air is supplied continuously through a filter chain consisting of silica gel, activated charcoal, and silica gel, in that order. This air is warmed to the same temperature as the stimulus bottles, and led to two funnels, not shown in the diagram, which serve respectively to provide a pure air replacement for odorous air drawn off from the stimulus bottles, and a source of

pure air for the stimulus mixture.

Thresholds are found by metering various amounts of odorous air into the syringe by proper adjustment of the stopcock, C, and then filling the syringe with pure air. This provides a very fine control of concentration and, since a knowledge of the vapor pressure of the odorous material, its molecular weight, the molecular weight of the oil, and the temperature, permit calculation of the concentration in the bottle, the stimulus concentration may be stated in molecular terms. There is no evidence of progressive threshold shifts with continuing drawing off of odorous air, and so it is assumed that evaporation is rapid enough to prevent more than negligible dilution during a series of trials.

A fairly constant environment is provided for O by a small temperature-controlled chamber in which he sits. This chamber is kept at 20°C, and all air therein is filtered through charcoal filters. It is glass-lined, except for zinc holding strips and a few stainless steel inserts. O's chair is covered with a polyethylene sack. This chamber provides a constant, but not perfectly "baseline" environment. Constancy is aided to a considerable degree by the fact that the chamber is in a room provided with a temperature and humidity controlled air supply from a central ventilating system. Temperature variation in the chamber is very slight -- too small to be measured with an ordinary laboratory thermometer.

The experimental routine for obtaining a single threshold is as follows: After all connections have been made, a small amount of odorous air is drawn into the syringe, and then the syringe is filled with pure air by retracting the plunger while the nosepiece is placed in the pure-air funnel. Now O places the nosepiece in his nostril, and the blast is delivered

by switching on the motor. After every trial, the rack and plunger are withdrawn after lifting the motor a small amount on its hinged mounting, and while the nosepiece is in place in the pure-air funnel. Several purging strokes of the plunger are then given by hand, and another trial begun. In actual practice, this proceeds very rapidly -- as rapidly, indeed, as trials should be given, since adaptation must be avoided. Between each pair of O's the nosepieces are washed and sterilized with ultraviolet light.

Sample results from four Os are available. In order to test the accuracy of the apparatus, it was decided to use two dilutions of two substances. Thus, it is possible to decide if the dilution in mineral oil and the metering provided by the syringe yield comparable results. That is, even if the concentration of odorous molecules in the absorption bottles differs, the thresholds in molecular terms for the same substance should remain the same. The two substances used were methyl salicylate and benzene.[4] There were unexpectedly large individual differences in sensitivity to methyl salicylate, and cross-comparison was not possible except for one inexperienced O. For benzene, however, comparison of two levels of dilution was made for all four O's. These results, each threshold

---

Insert Table I about here

---

being an average of three ascending determinations, are given in Table I. Correspondence is quite close, especially considering the small number of trials. It should be pointed out that these thresholds are recognition

thresholds, which we have found to be easier to judge than the absolute thresholds. Shown also in Table I are the thresholds for methyl salicylate.

The method of calculating thresholds in terms of molar ratios may be of interest. First of all, one must know the vapor pressure of the odorous substance. The best source is probably Timmermans.[5] If only the boiling point is known, it is possible to estimate the vapor pressure (as indeed was done for methyl salicylate in calculating the thresholds in this note).[6] Now, from the densities and the molecular weights of the odor and the mineral oil, one can find the molar ratio of the solution, and hence calculate the vapor pressure of the substance as diluted (the vapor pressure of the mineral oil is very low, and is neglected), by multiplying the vapor pressure by the mole fraction. The molar ratio of odor in the absorption bottle is then determined by reference to the air pressure, and finally, the molar concentration of the threshold blast is found from the volume of odorous air in the total mixture from which the blast is delivered.

Since the actual number of odorous molecules reaching the olfactory epithelium must depend upon a variety of factors, few of them the same for any two methods, no guess at the number of molecules involved in a barely supra-threshold stimulus will be attempted here, nor do comparisons with thresholds obtained by other methods have much meaning. If an apparatus can provide stable and homogeneous conditions with which to work, and if no variables other than stimulus concentration, stimulus substance, and O determine the thresholds, it has fulfilled its function. It appears that the apparatus herein described meets these criteria.

FOOTNOTES

*The work reported herein was performed under Contract No. N6onr-27515, Project NR140-008 between the University of California and the Office of Naval Research

1. Bernice M. Wenzel, Techniques in olfactometry: a critical review of the last one hundred years. Psychol. Bull., 1949, 45, 231-247.

2. F. N. Jones, A test of the Elsberg technique of olfactometry, This Journal. In press (January, 1953); J. A. Whittenburg, The effects of general activity and extended diurnal variation on olfactory sensitivity. Technical Report No. 11, Project DA-49-007-MD-222(O.I. 19-52), Army Medical Research and Development Board.

3. Obtained from the Connecticut Hard Rubber Company, New Haven.

4. Obtained respectively from the General Chemical Company and the Eastman Kodak Company.

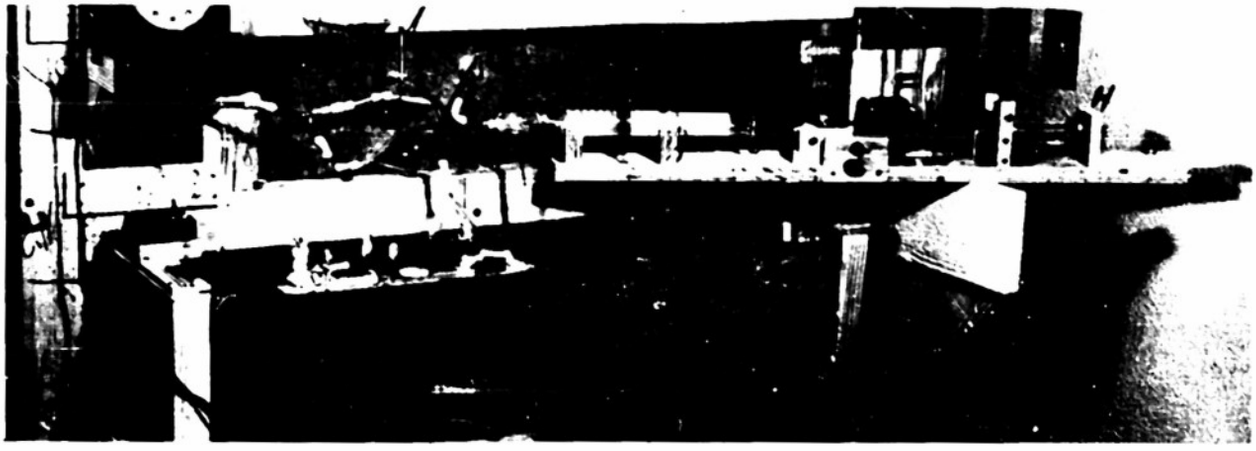
5. Jean Timmermans, Physico-Chemical Constants of Pure Organic Compounds. Elsevier, New York. 1950.

6. G. W. Thomson, Determination of Vapor Pressure. Chapt. V in Arnold Weissberger (Ed.), Technique of Organic Chemistry. Vol. I, Part 1, Physical Methods of Organic Chemistry. Interscience, New York, 1949. Formula 97, p. 228 is the one used here.

Table I

Thresholds for Benzene and Methyl Salicylate,
in Molar Ratios

O	Benzene 0.2%	0.4%	Methyl Salicylate
AB	1.7×10^{-4}	1.9×10^{-4}	4.1×10^{-7}
SH	1.6×10^{-4}	1.8×10^{-4}	3.4×10^{-6}
FMJ	1.8×10^{-4}	1.5×10^{-4}	3.6×10^{-6}
MHJ	1.1×10^{-4}	1.3×10^{-4}	3.6×10^{-8}
Av.	1.5×10^{-4}	1.6×10^{-4}	2.6×10^{-6}



A

Figure 1

Diagram of the Essential Portions of the Apparatus